REMARKS

Amended Claims are Patentable

Unique methods of utilizing transcription factor decoys which have the same sequence as Shear Stress Response Elements (SSRE's) are claimed. The methods described provide several different ways to monitor changing cellular expression in response to shear stress and/or use of transcription factor decoys to alter cellular response to shear stress. We have carefully considered the recommended changes and amended the current specification and claims. We believe the following amendments place the pending claims in condition for allowance, and no new matter is added to the specification.

The term "directed against" in the specification identified transcription factor decoy sequences which bind a transcription factor. The term "directed against" was removed from the pending claims. Removal of the phrase "directed against" merely removes the terminology and is not intended to reflect changes in the scope of the claims.

Claim 1 contains all of the limitations of previously dependent and allowable claim 4 (OA 6/11/01). Claim 6 correctly depends from claim 1 and is placed in condition for allowance (OA 6/11/01 and OA 9/23/01).

Claim 10 "wherein the concentration of said oligonucleotide sequence is from about 10 nM to about 10 mM," found in the original specification, was corrected in the first Office Action. Claim 10 derives support from the original claims and from the specification as demonstrated on page 10, line 4 of the application. The typographical error on page 10 of "m" (meters) for "M" (Molar) was corrected and a substitute sheet is included.

Claims 1-3, 5, 7-10, and 27 are directed to a method of culturing cells which mimic cells found *in vivo*. Using the methods described in this application, one of ordinary skill in the art would be able to culture cells which differentiate to form structures and expression patterns, such as renal islets, which are similar to cells isolated from tissues. The use of

transcription factor decoys allows the scientist to modulate shear stress response as demonstrated in the specification and distinctly claimed. Ando et al. (Jpn Heart J., Vol. 37, No. 1, Jan. 1996, p 19-32) speculate on the sequence and importance of an SSRE, but do not teach the use of SSRE sequences for modulating transcription factor activity. The embodiment of this claim is a method of using SSRE's to modulate transcription factor activity. The use of a single ODN to deliver and modulate transcription factor activity is novel. The specification has demonstrated that a single ODN containing an SSRE and the complement thereof provides a method of modulating MnSOD, ICAM, megalin, cubulin, erythropoietin, and 1-a-hydroxylase expression. The SSRE can also be used to modulate the levels of the 1,25-dihydroxy-vitamin D3 biomolecule. The specification clearly states that the SSRE regulates "genes specific for renal proximal tubular epithelial cells, including megalin, cubulin, the extracellular calcium sensing receptor, and the microvillar structural protein villin" (Application 09/532,001, page 21, line 3).

Transcription factor decoy sequences have been described in Dzau (WO199511687, published 5/4/95) and demonstrate the level of art at the time. Dzau et al. describe double-stranded ODN's which are delivered through liposomal vectors and injection, but do not describe a plausible method for large scale delivery of a transcription factor decoy. The use of single-stranded ODN's for delivery of the SSRE and the complement provide a vehicle for rapid and efficient delivery of SSRE's to the nucleus as demonstrated in the specification.

Kachician and associates (J. Clin. Invest., Vol. 96, Aug. 1995, p 1169-1175) perform two experiments which test the binding of SSRE's by NF-κB. The first experiment included gel retardation assays and DNAse footprinting of NF-κB complexes with SSRE elements with variable sequences. The second experiment involved the use of SSRE promoter elements to monitor reporter gene (CAT not a shear induced protein) expression under various stress conditions. Neither involved a single-stranded ODN or nuclear delivery of a

transcription factor decoy. The use of a single-stranded ODN to deliver a SSRE in its entirety, both coding sequence and complement, provides a novel method of altering transcription factor activity.

Applicants request consideration of the claims as amended and allowance of all pending claims.

The requisite fees, if any, are submitted herewith. No other fees are believed to be due for this submission. However, should there be any additional fees required, please charge such additional fees to Deposit Account No. 14-0116.

Respectfully submitted,

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